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Remarks

Further and favorable reconsideration is respectfully requested in view of the foregoing amendments and following remarks.

Thus, in response to the first two paragraphs on page 2 of the Office Action, the specification, abstract and claims have been amended to correct additional errors, including those mentioned by the Examiner.

In addition, the claims have been amended in response to the rejection of claims 21-38 under the second paragraph of 35 U.S.C. § 112, rendering all grounds for this rejection moot, with the single exception of the Examiner's comments concerning claim 30. This ground of rejection, relating to claim 30, is respectfully traversed.

The Examiner states that it is unclear that mere inoculation in step (ii) will result in conversion to gypsum. The Examiner then questions whether this is a chemical step or a microbial step, and whether or not the concentration is microbial.

In response to this, please note that the cyanobacteria absorb the calcium salts present and by internal metabolism convert the salts to free calcium ions. Similarly, the cyanobacteria also produce sulphate ions by their internal metabolism. The calcium and sulphate ions then react with each other to form gypsum, which precipitates. It is not necessary to separately add chemicals for the formation and separation of the gypsum.


The formation and precipitation of gypsum can be commonly seen in salt ponds where both calcium and sulphate salts are independently present.

Reference can also be made to the following web page (copy enclosed) for more details in this regard. <http://www2.cwr.uwa.edu.au/~segal/>

Therefore, in view of the foregoing amendments and remarks, it is submitted that the application is in condition for allowance. Such allowance is solicited.

Respectfully submitted,

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Project Title

Algal polysaccharide and primary production in solar salt ponds.

Project Summary

Polysaccharide production by benthic algae interferes with salt crystal structure and efficiency

of commercial salt production. There are no quantitative data on the role of this process in salt production and only anecdotal evidence that increased brine viscosity from polysaccharides leads to defective salt crystal formation. This project will conceptualise and quantify the environmental processes leading to polysaccharide formation through identification of the relevant benthic algae, assessment of their response to environmental conditions and measurement of polysaccharide excretion rates. This information will be used to optimise salt production in the Useless Loop solar salt ponds in Western Australia.

Study Area

Useless Loop is located in Shark Bay, Western Australia approximately 700 km north of Perth. The salt production facility is split into two series of ponds at Useless Inlet and Useless Loop (Figure 1), where seawater is continuously evaporated along the series of ponds until sodium chloride crystallises.



Figure 1 Location of Useless Inlet and Useless Loop. (Stenbeck *et al.*, 1999) ([full image](#))

The primary ponds are located at Useless Inlet and the secondary concentrators and the crystallisers are located at Useless Loop. The two series of ponds are connected by a 20 km flume. The salt crystals form on the floor of the final Useless Loop crystallisers, where it is then harvested (Figure 2). Shark Bay Salt Joint Venture produces around 1 million tonnes of salt (NaCl) per year. The ponds of the primary system are larger, with the first pond PM1 2500 ha in area. The primary system ponds generally decrease in depth and area down to 200-300 ha before entering the plume. The secondary concentrators and crystallisers are considerably smaller at 20-80 ha. The ratio of area of the primary to secondary ponds is generally 10 to 1 in solar salt works.

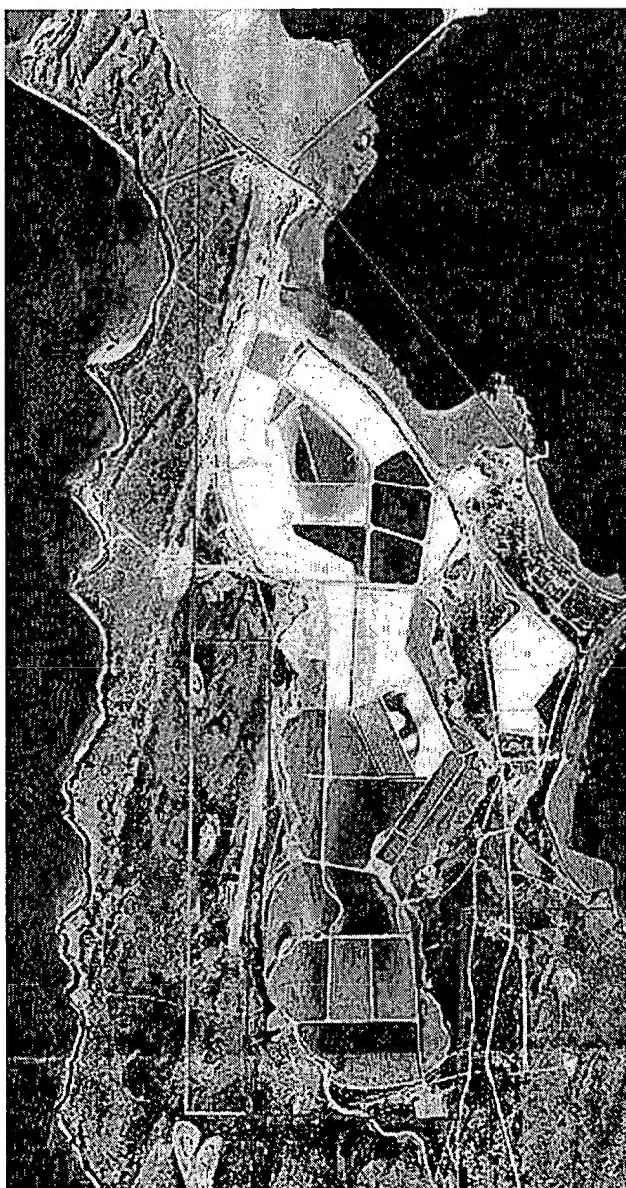


Figure 2 Useless Loop ponds - salt is harvested in the light-coloured ponds.

The multi-pond system presents an opportunity to investigate the changes in primary production, the fate of nutrients and nutrient limitation of production along the salinity gradient. It is anticipated that as the salinity increases, respiration will become greater than photosynthesis and that benthic algal production will become dominant. Properly functioning ponds strip nutrients from the water column, helping to reduce algal production as salinity increases, improving salt production and quality. Generally, marine waters are nitrogen limited and freshwater lakes are phosphorus limited. The input of seawater would suggest that the ponds would be nitrogen limited but with the salinities present in the ponds much greater than that of seawater, it will be of interest to see if this is the case.

The ponds located at the end of the primary system at Useless Inlet (P3) have been identified as the major contributors to polysaccharide production. The polysaccharide slime is known to dissolve into the brine, increasing the dissolved organic carbon concentration and brine viscosity. Both can be detrimental to salt production and crystal quality (Davis and Giordano, 1996). The benthic gypsum and cyanobacterial mats are well established and extensive in these ponds. Nutrient bioassays will be conducted on these mats to determine the conditions favourable for polysaccharide production.

The salinities of the ponds present harsh conditions for the biota, from marine (35ppt) up to precipitation of sodium chloride (350ppt). In between the two extremes, calcium carbonate (60ppt) and calcium sulphate (gypsum, 110ppt) also precipitate, forming benthic crusts (Figure 3). As the salinity increases, the ionic strength of the water increases and as a result the diversity decreases (Javor, 1989). Fish are known to exist in the Shark Bay ponds but only until around 80ppt. Macroalgae, seagrasses and phytoplankton also disappear at around 80-100ppt if the ponds are operating correctly. The organisms which remain include cyanobacteria, diatoms, photosynthetic bacteria, halobacteria and brine shrimp (*Artemia*). The cyanobacteria form mats which are made up of often quite distinct layers. Cyanobacteria and diatoms occupy the surface layers, with coloured and colourless sulphur bacteria underneath. A black sulphur-rich layer is often found at greater depths. In the gypsum ponds, the layers are quite distinct (Figure 3), but in the less saline ponds the layers are more compressed (<5mm). After the precipitation of sodium chloride, the magnesium concentration becomes so high that the bitters is almost sterile (Javor, 1989).

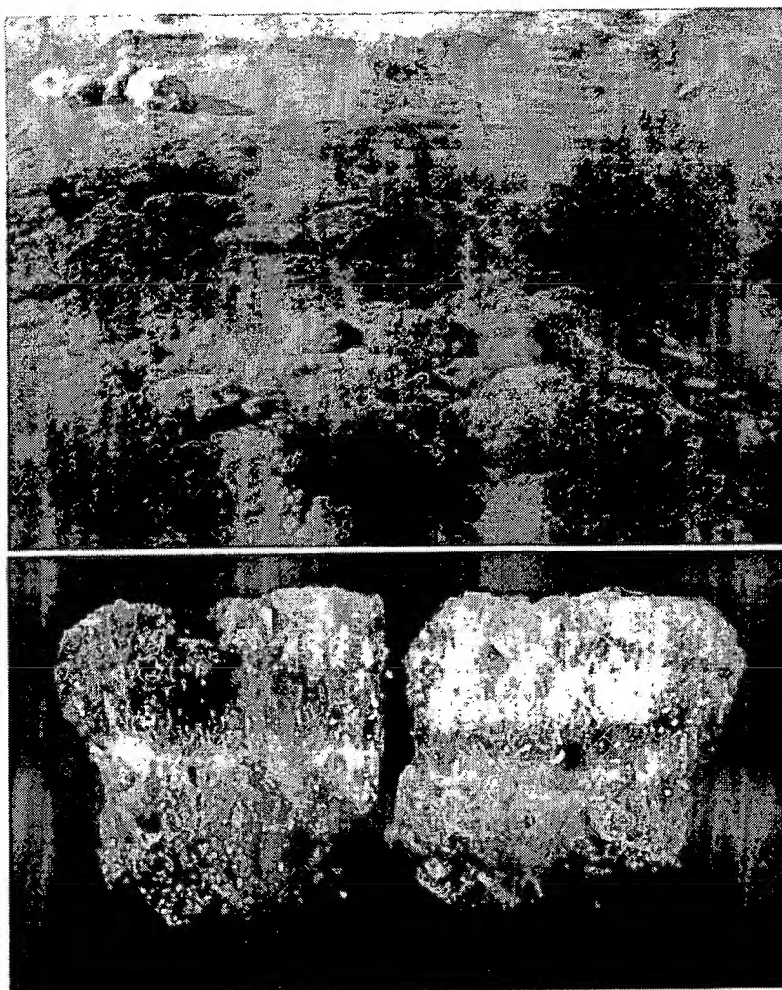


Figure 3 Gypsum-covered bottom of a salt pond (upper panel) and a cross-section of a piece of the gypsum crust showing layered microbial communities (lower panel) (Eilat, Israel). (Oren *et al.*, 1995)

The PhD proposal was submitted and accepted in April 2002. Field work is set to commence in June 2002.

Progress Updates and Results

Click here to view the [research proposal](#)

Click here to view a HTML version of the [proposal defence](#) presented 10/4/02

Click here to view a [summary](#) of the Winter 2002 field trip results

Click here to view a HTML version of the [Aquatic Chemistry and Ecology Seminar](#) presented 4/12/02

Further Information

International Society for Salt Lake Research

Australian Society for Limnology

Davis, J.S. and Giordano, M. 1996. Biological and physical events involved in the origin, effects and control of organic matter in solar saltworks. *International Journal of Salt Lake Research* 4(4) 335-347.

Javor, B. 1989 *Hypersaline Environments - microbiology and biogeography*. Springer-Verlag Berlin, Germany

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Stal, L.J. and Caumette, P. (eds) 1994 *Microbial Mats- structure, development and environmental significance*. Springer-Verlag Berlin, Germany

Stenbeck, D.R., Horn, D.A. and Imberger, J. 1999 Optimisation of salt production in the Shark Bay salt fields - feasibility report . WP1476DS CWR, UWA Perth Western Australia

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